

## Improved the Shelf Life of Guava Fruits by Salicylic Acid against Postharvest Black Spot Disease

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### ABSTRACT

Guava fruit (*Psidium guajava* L.) is one of the most important popular fruits in the tropical and subtropical zones. It is a perishable and has a short postharvest shelf life at environment temperature due to climacteric ripening with changes in physicochemical properties; However, the most serious microorganisms attacked guava fruits are the fungi. Guava black spot (GBS) is a latent infection disease caused by the ascomycete fungal *Phyllosticta capitalensis*, that infect immature guava in the field. This postharvest decay can lead to significant economic losses. Since the commercial application of fungicides to control postharvest diseases found harmful to human and the ecosystems. Therefore, the search of finding alternative fungicides, safety and environmentally friendly strategy is a major aim of the researchers worldwide. Salicylic acid (SA) is one of the safe, natural compounds applied in the postharvest treatment of fruit. The present investigation aimed to evaluate the effect of different concentrations of SA on fungal growth in vitro and postharvest treatment of infected fruit to improve the shelf life of guava fruits. Guava fruit collected from local supermarket and measurement of disease incidence and severity (DI & DS%) carried out in the three seasons (2015-2017). SA at different concentrations assessed on mycelial linear growth inhibition (MLGI%) of *P. capitalensis* in vitro. Exogenous postharvest treatment of guava fruit with SA tested at five concentrations, three times of immersion and kept for three periods time of shelf life. DS% evaluated after three period time. Total soluble solids (TSS) and weight loss (WL) evaluated after three period time. SA concentrations at 6 mmol/L significant on MLGI of *P. capitalensis* in vitro. Postharvest treatment with SA at 3 mmol/L significantly reduced DS% on guava fruit, at three immersion time and after three periods of shelf life, compared to control. SA with a concentration at 4 mmol/L decreased TSS and WL after three periods of shelf life. Postharvest treatment with SA solutions had an overall positive effect on fruit quality of guava during shelf life. A decay incidence decreased, TSS and weight loss noticed in treated guava fruit compared to control. The present study demonstrated the efficacy and potential of SA solutions in preserving the shelf life of a highly perishable like guava fruits.

**Keywords:** Guava black spot, *Phyllosticta capitalensis*, postharvest disease, Salicylic acid

### INTRODUCTION

Guava fruit (*Psidium guajava* L., Family Myrtales) is an important and the most popular commercial in Egypt, tropical and subtropical worldwide (Batista Silva *et al.*, 2018). The cultivated areas in Egypt are about 47.620 feddans with an annual production 350 thousand tons of fruits (Anonymous, 2016). It is an excellent source of phosphorus, vitamin C (Murmu & Mishra, 2018a, 2018b). Guava fruit is a climacteric and perishable after harvest (Bashir & Abu-Goukh, 2003; D. Singh & Sharma, 2018; S. Singh & Pal, 2008). The fungal pathogens on fruits controlled by low temperature storage. A case in point, guava fruit storage below 13°C causes chilling injury (CI) symptoms (Gonzalez-Aguilar, Tiznado-Hernandez, Zavaleta-Gatica, & Martinez-Tellez, 2004; Sevillano, Sanchez-Ballesta, Romojaro, & Flores, 2009). Losses due to postharvest fungal diseases may occur during postharvest handling, from harvest to consumption. The total production of fruits and vegetables produced globally, wasted between 15-50% at the postharvest stage by pathogen decay, with a high score in developing countries (Gustavsson, Cederberg, Sonesson, Van Otterdijk, & Meybeck, 2011). GBS disease caused by *P. capitalensis*, is the fungal disease causing severe postharvest a significant loss in quality of guava (Arafat, 2018). The symptoms of GBS appear of the infected fruit were small, slightly sunken on mature fruits. Application of fungicide treatments even now is still one of the most efficient methods to reduce postharvest decay. However, the commercial application of fungicides to control postharvest diseases found harmful to human and the environment. Consequently, strongly needs to discovering alternative fungicides, safety and environmentally friendly strategy is a major aim of the researchers worldwide. In the last few decades, there has been an increasing interest in chemical compounds with low toxicity and generally recognized as safe (GRAS)

compounds for the control of postharvest fruit diseases to reduce fungicides use to a minimum (Burdock & Carabin, 2004). There is a natural occurrence of induced resistance (IR) to protect fruits from the disease. Elicitors such as plant hormones are compounds, which activate chemical defense in fruits. Varying biosynthetic pathways activated in treated fruits depending on the compound used. There are many published studies (Ali, Hahn, & Paek, 2007; Cajuste & Lafuente, 2007; Gonzalez-Aguilar *et al.*, 2004; Heil & Bostock, 2002; Lattanzio, Lattanzio, & Cardinali, 2006; Ruiz-García & Gómez-Plaza, 2013; Siboz, Bertling, & Odindo, 2014; Zabka & Pavela, 2013) that describe the importance of phenolic compounds, which naturally occur in plants and the role in protecting from both biotic and abiotic stresses. Extensive research has shown that, SA (Ortho-hydroxybenzoic acid), and its derivatives, which generated by plants as part of their defense systems against pathogen attack and environmental stress. Moreover, it is one of the safe, natural compounds used in postharvest processing of fruit. While abiotic stress factors may alter the endogenous SA levels in the plants and therefore, elicit several defense mechanisms in plants, more the latest research studies have focused on the effects of the exogenous application of SA (Wills & Golding, 2016). Recent research has found; the postharvest exogenous application of SA improves the efficacy of the plant defense mechanisms under both biotic and abiotic stresses. In the past two decades, studies have increased our understanding of Salicylic acid application to plants (Supapvanich & Promyou, 2013). It is widely recognized that SA reduces the respiratory rate (Asghari & Aghdam, 2010) inhibits an ethylene biosynthesis (F. Xu, Liu, Xu, & Fu, 2019), induces the expression of defense genes (Meena, Marimuthu, & Velazhahan, 2001; Wen *et al.*, 2005) and decreases lipid oxidation and membrane senescence (M. Kazemi, M. Aran, & S. Zamani, 2011;

Kazemi, Hadavi, & Hekmati, 2011) in plants. Recent evidence suggests that, SA as postharvest treatment is limited to concentrations that are safe at plants, with an optimum range of about 0.5-2mM (Babalar, Asghari, Talaei, & Khosroshahi, 2007). Moreover, SA is looked at an essential signaling molecule, which plays an important role in regulating disease resistance and reducing the production rate of superoxide anions in fruits (Ding & Wang, 2003; Horváth, Szalai, & Janda, 2007), such as kiwi (Fatemi, Mohammadi, & Aminifard, 2013), cherry (Dokhanieh, Aghdam, Fard, & Hassanpour, 2013), apple (da Rocha Neto, Luiz, Maraschin, & Di Piero, 2016; Mo *et al.*, 2008), tomato (Aghdam, Asghari, Khorsandi, & Mohayjeji, 2014), mango (Damodaram *et al.*, 2015) and pear (Wang & Chen, 2010). Satisfactory SA results have been reported for control of *Penicillium expansum* in the sweet cherry (X. Xu & Tian, 2008), grey mold in peach (Zhang *et al.*, 2008) and fungal decay in persimmon fruit (Khademi, Zamani, Mostofi, Kalantari, & Ahmadi, 2012). The present investigation aimed to evaluate the effect of different concentrations of SA on fungal growth *in vitro* and postharvest treatment of infected fruit to improve the shelf life of guava fruits.

## MATERIALS AND METHODS

### Plant materials:

One hundred samples for each three seasons (2015-2017) of guava fruit (*Psidium guajava* L.) cv. White Balady, randomly collected from different local markets in El-Kharga city (25.4390 N, 30.5586 E), New Valley governorate, Egypt. The collected samples kept in sterilized polyethylene bags and brought to the laboratory of the Plant Pathology Department, Faculty of Agriculture, New Valley University, Egypt.

### Measurement of percent disease incidence (%DI):

The percent disease incidence (%DI) measured and calculated for each season based on the formula:

$$\% DI = \frac{\text{Number of infected fruits}}{\text{total number of fruits}} \times 100$$

### Measurement of percent disease severity (%DS):

The result recorded using the decay index (Di) and severity (%DS) for each season as follows:

$$Di = \frac{\text{Sum (number of fruits per category} \times \text{category number)}}{\text{total number of fruits infected}}$$

$$\% DS = \frac{Di}{4} \times 100$$

### Pathogenic fungal:

The pure culture of isolate *P. capitalensis* (caused GBS disease) obtained from the Plant Pathology Department-Faculty of Agriculture- New Valley University. The pathogenic fungal identified by molecular analysis as *P. capitalensis*, here represented by the code (ARAFAT-GF5), according to the GenBank (Accession number- LC269950.1; GI: 119461242) with a synonym: *Guignardia mangiferae* (Arafat, 2018).

### Effect of SA at different concentrations on (%) mycelial linear growth inhibition (MLGI) of *Phyllosticta capitalensis* *in vitro*:

Antifungal of SA activity determined by poisoned food technique to evaluate the antifungal effect against pathogenic fungal (Khademi *et al.*, 2012; X. Xu & Tian, 2008; Zhang *et al.*, 2008). SA tested at concentration (0, 1, 2, 3, 4, 5 and 6 mmol/L). Inoculum of the pathogenic fungal prepared by choice 5mm disc cut with a sterile cork-borer from 15-day old culture grown on PDA medium. The fungal

discs put on the PDA plate. The agar plates prepared by adding preferred concentration of SA at a temperature of 45-50°C. The plates then incubated at temperature 25°C for fungal. Each concentration replicated three times, and the experiment repeated two times. Colony diameter recorded by measuring the two-opposite circumference of the colony growth. Percentage inhibition of mycelial growth evaluated by comparing the colony diameter of poisoned plate (with SA) and control plate (without SA) and calculated using the formula as the follows:

$$\% IZ = \frac{DGc - DGt}{DGc} \times 100$$

Where IZ= Percent of the inhibition zone, DGc= is the diameter of growth in control plate, and DGt= is the diameter of growth of the plate containing tested of SA.

### Effect of SA immersion on fruit disease severity%:

Guava fruits randomly distributed into three groups of 225 fruits per group for each of the shelf-life three periods (5, 10 and 15 days). Fruit of each group immersion into solutions of SA at five concentration 0, 1, 2, 3 and 4 mmol/L for three time 5, 10 and 15 min, respectively. Fruit allowed to dry for 30 min at room temperature and inoculated with a mycelial plug (5mm) of pathogenic fungal (Arafat, 2018). The fruits kept at room temperature 25±2 for three shelf-life time (5, 10 and 15 days). The percent of the DS calculated as mentioned above. Each concentration contained three replicates of 15 fruits, and the experiment performed twice.

### Effect of SA immersion on fruit Total soluble solids (°Brix):

The TSS content of guava fruit juice, inoculated and healthy at three shelf life time, measured by using a hand refractometer 0-32 °Brix (Atago Co., Tokyo, Japan).

**Effect of SA immersion on fruit Weight loss (WL):** The percent weight loss of guava fruit inoculated and healthy at different concentrations of SA with three shelf life time, calculated as followed:

$$\% WL = \frac{Wp1 - Wu2}{Wp1} \times 100$$

While WL= is the percent of weight loss, Wp1= is the preliminary weight of the guava fruit, Wu2= is the ultimate weight of the guava fruit after shelf life days of the study.

### Statistical analysis:

Data from the effect of SA on MLGI% analyzed using one-way analysis of variance (ANOVA) and the Duncan's test. Data from the effect of SA on DS%, TSS and WL% subjected to two-way analysis of variance (ANOVA) by using CoHort Software, CA, USA to compare the means among the treatments and among the different time intervals. The follow-up of ANOVA includes with complete block randomizes design. The means of all treatments compared using Duncan's multiple rang test at  $P < 0.05$  as significant. Different data recorded and expressed as means ± S.E (Gomez, Gomez, & Gomez, 1984).

## RESULTS

### Measurement of %DI and %DS:

The results, as shown in Table 1, according to a survey for three seasons (2015-2017) for GBS disease of guava fruit, data show that, highest percentage of both DI% and DS% recorded in season 2017 (15 and 60%), respectively. Whereas, DI% and DS% recorded in both seasons 2015-2016 (10 and 12%) and (44 and 52%), respectively.

**Table 1. Measurement of DI and DS% in three seasons (2015-2017)**

Season	DI%	Decay Index	DS%
2015	10.00	2.2	44.00
2016	12.00	2.6	52.00
2017	15.00	3.0	60.00

**Effect of SA on (MLGI%) of *Phyllosticta capitalensis* in vitro:**

Table 2 provides the experimental data for the effect of SA on pathogenic fungal at different concentrations. More distant statistical tests revealed that, all concentrations of SA positive significant on mycelial growth of *P. capitalensis*. Further analysis showed that, all concentrations of SA positive to MLGI of *P. capitalensis*. The most effective treatment of SA concentration on linear growth recorded at 6 mmol/L with inhibition (100%), followed by 5, 4, 3, 2 and 1 mmol/L with inhibition (73.70, 62.81, 38.74, 25.26 and 15.58%), respectively. Moreover, control treatment recorded with inhibition (0%).

**Table 2. Effect of SA at different concentrations on Mycelial linear growth inhibition (%) of *P. capitalensis***

SA Concentrations (mmol/ L)	MLGI %
0	0 <sup>g</sup>
1	15.58 ± 0.316 <sup>t</sup>
2	25.26 ± 0.398 <sup>e</sup>
3	38.74 ± 0.357 <sup>d</sup>
4	62.81 ± 0.341 <sup>c</sup>
5	73.70 ± 0.208 <sup>b</sup>
6	100 <sup>a</sup>

The superscript letters indicated significantly between effect of concentrations.

**Effect of SA immersion on fruit disease severity%:**

**Table 3. Effect of salicylic acid at different concentrations on fruit decay after 5, 10 and 15 days**

SA conc.	Disease severity (%)							
	Shelf life after 5 days							
	Immersion Time						Mean	
	5 min		10 min		15 min		Decay Index	DS%
Decay Index	DS%	Decay Index	DS%	Decay Index	DS%			
0	1.45±0.013	29.00 <sup>a</sup>	1.47±0.021	29.40 <sup>b</sup>	1.48±0.020	29.60 <sup>c</sup>	1.47±0.010	29.60 <sup>a</sup>
1	1.39±0.021	27.80 <sup>a</sup>	1.35±0.017	27.00 <sup>b</sup>	1.29±0.017	25.80 <sup>c</sup>	1.34±0.012	26.80 <sup>b</sup>
2	0.97±0.016	19.40 <sup>a</sup>	0.94±0.016	18.80 <sup>b</sup>	0.82±0.022	16.40 <sup>c</sup>	0.91±0.014	18.20 <sup>c</sup>
3	0.00	0.00 <sup>a</sup>	0.00	0.00 <sup>b</sup>	0.00	0.00 <sup>c</sup>	0.00	0.00 <sup>c</sup>
4	0.73±0.015	14.6 <sup>a</sup>	0.56±0.016	11.20 <sup>b</sup>	0.51±0.019	10.20 <sup>c</sup>	0.60±0.017	12.00 <sup>d</sup>
Mean	0.91±0.062	18.2 <sup>a</sup>	0.86±0.063	17.20 <sup>b</sup>	0.82±0.062	16.40 <sup>c</sup>		
	Shelf life after 10 days							
0	2.55±0.013	51.00 <sup>a</sup>	2.57±0.012	51.40 <sup>b</sup>	2.55±0.017	51.00 <sup>c</sup>	2.56±0.008	51.20 <sup>a</sup>
1	1.85±0.013	37.00 <sup>a</sup>	1.67±0.018	33.40 <sup>b</sup>	1.63±0.016	32.60 <sup>c</sup>	1.72±0.017	34.40 <sup>b</sup>
2	1.49±0.018	29.80 <sup>a</sup>	1.49±0.019	29.80 <sup>b</sup>	1.37±0.016	27.40 <sup>c</sup>	1.45±0.013	29.00 <sup>c</sup>
3	0.00	0.00 <sup>a</sup>	0.00	0.00 <sup>b</sup>	0.00	0.00 <sup>c</sup>	0.00	0.00 <sup>c</sup>
4	1.25±0.013	25.00 <sup>a</sup>	1.22±0.011	24.40 <sup>b</sup>	1.13±0.021	22.60 <sup>c</sup>	1.20±0.012	24.00 <sup>d</sup>
Mean	1.43±0.097	28.60 <sup>a</sup>	1.39±0.097	27.80 <sup>b</sup>	1.34±0.096	26.80 <sup>c</sup>		
	Shelf life after 15 days							
0	4.17±0.021	83.40 <sup>a</sup>	4.23±0.019	84.60 <sup>b</sup>	4.22±0.023	84.40 <sup>c</sup>	4.30±0.013	86.00 <sup>a</sup>
1	3.54±0.013	70.80 <sup>a</sup>	3.43±0.013	68.60 <sup>b</sup>	3.33±0.013	66.60 <sup>c</sup>	3.44±0.015	68.80 <sup>b</sup>
2	3.25±0.013	65.00 <sup>a</sup>	3.15±0.017	63.00 <sup>b</sup>	3.03±0.013	60.60 <sup>c</sup>	3.14±0.015	62.80 <sup>c</sup>
3	0.00	0.00 <sup>a</sup>	0.00	0.00 <sup>b</sup>	0.00	0.00 <sup>c</sup>	0.00	0.00 <sup>c</sup>
4	2.91±0.018	58.20 <sup>a</sup>	2.55±0.013	51.00 <sup>b</sup>	2.44±0.013	48.80 <sup>c</sup>	2.63±0.031	52.60 <sup>d</sup>
Mean	2.77±0.168	55.40 <sup>a</sup>	2.67±0.168	53.40 <sup>b</sup>	2.61±0.166	52.20 <sup>c</sup>		

The interaction between DS%, immersion time and SA concentrations treatment mean in a column are significantly different at  $P < 0.05$  (Duncan's multiple rang test). Means followed by the same letter do not differ significantly. Each value represents mean and ± SE.

**Effect of SA immersion on fruit TSS (°Brix):**

The Total soluble solids of guava fruit determined and summarized in Table 4. The results show that, all concentrations of SA tested effect on TSS. After 5 days of shelf life, TSS of guava fruit treatment with SA concentration at 1 and 2 mmol/L mean recorded (5.90 and

The purpose of experiment 3 was to decreased fruit decay and increased shelf life period by postharvest treatment of guava fruits with salicylic acid at different concentrations and immersion time. The differences of DS% after 5, 10 and 15 days highlighted in (Table 3). Based on the results, the highest decreased of guava fruits DS% after 5 days of shelf life, with SA at concentration 3 mmol/L mean recorded (0% DS). Followed by 4 mmol/L but with browning on surface fruit, mean recorded (12% DS). While, the latest reduced of DS% with SA at 2, 1 and 0 mmol/L mean recorded (18.20, 26.80 and 29.60% DS), respectively. The most effective time for immersion fruit with SA to decreased DS% for 15 min, followed by 10 and 5 min mean recorded (16.40, 17.20 and 18.20% DS), respectively. After 10 days of shelf life, DS% of guava fruit treatment with SA concentration at 3 mmol/L mean recorded (0% DS). Followed by 4 mmol/L but with browning on surface fruit, mean recorded (24% DS). While, the latest reduced of DS% with SA at 2, 1 and 0 mmol/L mean recorded (29.00, 34.40 and 51.20% DS), respectively. The most effective immersion time of fruit with SA to decreased DS% for 15 min, followed by 10 and 5 min mean recorded (26.80, 27.80 and 28.60% DS), respectively. After 15 days of shelf life, DS% of guava fruit treatment with SA concentration at 3 mmol/L mean recorded (0% DS). Followed by 4 mmol/L but with browning on surface fruit, mean recorded (52.60% DS). While, the latest reduced of DS% with SA at 2, 1 and 0 mmol/L mean recorded (62.80, 68.80 and 86.00% DS), respectively. The most effective immersion time of fruit with SA to decreased DS% for 15 min, followed by 10 and 5 min mean recorded (52.20, 53.40 and 55.40% DS), respectively.

5.90 °Brix), followed by concentration 3 mmol/L mean recorded (5.70 °Brix) and the latest concentration 4 mmol/L mean recorded (5.30 °Brix) compared to control mean which recorded (6.14 °Brix). Immersion time effect on TSS, for 5 min mean recorded (6.12 °Brix), while immersion time for 10 and 15 min mean recorded (6.04

and 5.99 °Brix) compared with zero time (5.10 °Brix). immersion time for 5 min found significant, followed by 10 and 15 min. No significant founded between immersion time 10 and 15 min. After 10 days of shelf life, TSS of guava fruit treatment with SA concentration at 1 mmol/L mean recorded (8.55 °Brix), followed by concentration 2, 3 and 4 mmol/L mean recorded (7.60, 6.90 and 5.87 °Brix), respectively. Compared with control mean recorded (8.93 °Brix). Immersion time effect on TSS, for 5, 10 and 15 min mean recorded (8.47, 8.41 and 8.31 °Brix), respectively.

No significant founded between immersion time for 5, 10 and 15 min. After 15 days of shelf life, TSS of guava fruit treatment with SA concentration at 1 mmol/L mean recorded (9.91 °Brix), followed by concentration 2, 3 and 4 mmol/L mean recorded (9.20, 8.38 and 7.68 °Brix), respectively. Compared with control mean recorded (10.58 °Brix). immersion time effect on TSS for 5, 10 and 15 min mean recorded (10.67, 10.47 and 10.36 °Brix), respectively. All immersion time for 5, 10 and 15 min founded significant, respectively.

**Table 4. Effect of salicylic acid treatments at different concentrations on fruit TSS (°Brix)**

SA Conc.	TSS (°Brix)				
	Shelf life after 5 days				
	Immersion Time				Mean
0 Time	5 min	10 min	15 min		
0	5.10±0.050 <sup>c</sup>	6.44±0.013 <sup>a</sup>	6.50±0.015 <sup>b</sup>	6.53±0.013 <sup>b</sup>	6.14±0.080 <sup>a</sup>
1	5.10±0.050 <sup>c</sup>	6.25±0.013 <sup>a</sup>	6.31±0.010 <sup>b</sup>	6.32±0.011 <sup>b</sup>	5.99±0.096 <sup>b</sup>
2	5.10±0.050 <sup>c</sup>	6.18±0.016 <sup>a</sup>	6.14±0.015 <sup>b</sup>	6.13±0.024 <sup>b</sup>	5.88±0.061 <sup>c</sup>
3	5.10±0.050 <sup>c</sup>	6.05±0.026 <sup>a</sup>	5.96±0.020 <sup>b</sup>	5.87±0.019 <sup>b</sup>	5.74±0.051 <sup>d</sup>
4	5.10±0.050 <sup>c</sup>	5.65±0.019 <sup>a</sup>	5.28±0.030 <sup>b</sup>	5.14±0.021 <sup>b</sup>	5.29±0.033 <sup>e</sup>
Mean	5.10±0.022 <sup>c</sup>	6.12±0.032 <sup>a</sup>	6.04±0.050 <sup>b</sup>	5.99±0.056 <sup>b</sup>	
Shelf life after 10 days					
0	5.10±0.050 <sup>b</sup>	10.17±0.040 <sup>a</sup>	10.20±0.035 <sup>a</sup>	10.24±0.016 <sup>a</sup>	8.93±0.288 <sup>a</sup>
1	5.10±0.050 <sup>b</sup>	9.75±0.029 <sup>a</sup>	9.70±0.011 <sup>a</sup>	9.62±0.021 <sup>a</sup>	8.55±0.260 <sup>b</sup>
2	5.10±0.050 <sup>b</sup>	8.53±0.027 <sup>a</sup>	8.47±0.017 <sup>a</sup>	8.31±0.017 <sup>a</sup>	7.60±0.189 <sup>c</sup>
3	5.10±0.050 <sup>b</sup>	7.64±0.013 <sup>a</sup>	7.48±0.016 <sup>a</sup>	7.40±0.033 <sup>a</sup>	6.90±0.137 <sup>d</sup>
4	5.10±0.050 <sup>b</sup>	6.75±0.034 <sup>a</sup>	6.16±0.016 <sup>a</sup>	5.96±0.026 <sup>a</sup>	5.87±0.061 <sup>e</sup>
Mean	5.10±0.022 <sup>b</sup>	8.47±0.166 <sup>a</sup>	8.41±0.171 <sup>a</sup>	8.31±0.179 <sup>a</sup>	
Shelf life after 15 days					
0	5.10±0.050 <sup>c</sup>	12.55±0.023 <sup>a</sup>	12.39±0.020 <sup>ab</sup>	12.28±0.026 <sup>b</sup>	10.58±0.412 <sup>a</sup>
1	5.10±0.050 <sup>c</sup>	11.59±0.015 <sup>a</sup>	11.51±0.024 <sup>ab</sup>	11.43±0.014 <sup>b</sup>	9.91±0.632 <sup>b</sup>
2	5.10±0.050 <sup>c</sup>	10.80±0.021 <sup>a</sup>	10.51±0.015 <sup>ab</sup>	10.38±0.018 <sup>b</sup>	9.20±0.309 <sup>c</sup>
3	5.10±0.050 <sup>c</sup>	9.65±0.019 <sup>a</sup>	9.43±0.017 <sup>ab</sup>	9.33±0.018 <sup>b</sup>	8.38±0.247 <sup>d</sup>
4	5.10±0.050 <sup>c</sup>	8.74±0.024 <sup>a</sup>	8.52±0.019 <sup>ab</sup>	8.36±0.019 <sup>b</sup>	7.68±0.195 <sup>e</sup>
Mean	5.10±0.022 <sup>c</sup>	10.67±0.157 <sup>a</sup>	10.47±0.126 <sup>ab</sup>	10.36±0.164 <sup>b</sup>	

The interaction between TSS, immersion time and SA concentrations treatment mean in a column are significantly different at  $P < 0.05$  (Duncan's multiple rang test). Means followed by the same letter do not differ significantly. Each value represents mean and ± SE.

**Effect of SA immersion on fruit Weight loss (WL):**

Guava fruits treated with SA at three concentrations and storage at ambient temperature for three time periods then the weight loss (WL) measured after 5, 10 and 15 days of fruit shelf live (Table 5). After 5 days of shelf life, the most effective concentration of SA to decreased WL% at 4 mmol/L mean recorded (7.6%) followed by 3, 2 and 1 mmol/L mean recorded (7.10, 7.13 and 7.19%) compared with control (7.28%). Immersion time effect on WL, the most effect time to decreased WL at 15 min mean recorded (7.14%), followed by 10 and 15 min mean recorded (7.15 and 7.17%). All immersion time founded significant. After 10 days of shelf life, the most effective concentration of SA to decreased WL% at 4 mmol/L mean recorded (7.14%) followed by 3, 2 and 1 mmol/L mean recorded (7.56, 8.13 and 8.46%) compared with control (9.26%). Immersion time effect on WL, the most effect time to decreased WL at 15 min mean recorded (8.02%), followed by 10 and 15 min mean recorded (8.11 and 8.19%). All immersion time founded significant. After 15 days of shelf life, the most effective concentration of SA to decreased WL% at 4 mmol/L mean recorded (10.01%) followed by 3, 2 and 1 mmol/L mean recorded (10.88, 12.21 and 13.39%) compared with control (14.94%). Immersion time effect on WL, the most effect time to decreased WL at 15 min mean recorded (12.10%), followed by 10 and 15 min mean recorded (12.23 and 12.53%).

**Table 5. Effect of salicylic acid treatments at different concentrations on fruit weight loss (%)**

Concentration	Weight losses (%)			
	Shelf life after 5 days			
	Immersion Time			Mean
5 min	10 min	15 min		
0	7.28±0.022 <sup>a</sup>	7.28±0.022 <sup>ab</sup>	7.28±0.022 <sup>b</sup>	7.28±0.013 <sup>a</sup>
1	7.21±0.022 <sup>a</sup>	7.21±0.010 <sup>ab</sup>	7.16±0.016 <sup>b</sup>	7.19±0.010 <sup>b</sup>
2	7.15±0.003 <sup>a</sup>	7.12±0.004 <sup>ab</sup>	7.12±0.011 <sup>b</sup>	7.13±0.005 <sup>c</sup>
3	7.10±0.007 <sup>a</sup>	7.10±0.007 <sup>ab</sup>	7.10±0.010 <sup>b</sup>	7.10±0.005 <sup>d</sup>
4	7.08±0.002 <sup>a</sup>	7.06±0.002 <sup>ab</sup>	7.04±0.002 <sup>b</sup>	7.06±0.003 <sup>e</sup>
Mean	7.17±0.011 <sup>a</sup>	7.15±0.011 <sup>ab</sup>	7.14±0.011 <sup>b</sup>	
Shelf life after 10 days				
0	9.26±0.003 <sup>a</sup>	9.26±0.003 <sup>b</sup>	9.26±0.003 <sup>c</sup>	9.26±0.002 <sup>a</sup>
1	8.51±0.018 <sup>a</sup>	8.49±0.019 <sup>b</sup>	8.39±0.017 <sup>c</sup>	8.46±0.013 <sup>b</sup>
2	8.22±0.022 <sup>a</sup>	8.12±0.017 <sup>b</sup>	8.05±0.017 <sup>c</sup>	8.13±0.015 <sup>c</sup>
3	7.75±0.029 <sup>a</sup>	7.56±0.016 <sup>b</sup>	7.36±0.013 <sup>c</sup>	7.56±0.023 <sup>d</sup>
4	7.24±0.012 <sup>a</sup>	7.13±0.013 <sup>b</sup>	7.05±0.013 <sup>c</sup>	7.14±0.013 <sup>c</sup>
Mean	8.19±0.080 <sup>a</sup>	8.11±0.086 <sup>b</sup>	8.02±0.090 <sup>c</sup>	
Shelf life after 15 days				
0	14.94±0.031 <sup>a</sup>	14.94±0.031 <sup>b</sup>	14.94±0.031 <sup>c</sup>	14.94±0.017 <sup>a</sup>
1	13.65±0.017 <sup>a</sup>	13.40±0.026 <sup>b</sup>	13.13±0.013 <sup>c</sup>	13.39±0.033 <sup>b</sup>
2	12.49±0.019 <sup>a</sup>	12.16±0.019 <sup>b</sup>	11.97±0.021 <sup>c</sup>	12.21±0.034 <sup>c</sup>
3	11.42±0.024 <sup>a</sup>	10.71±0.023 <sup>b</sup>	10.52±0.017 <sup>c</sup>	10.88±0.060 <sup>d</sup>
4	10.15±0.013 <sup>a</sup>	9.94±0.016 <sup>b</sup>	9.93±0.021 <sup>c</sup>	10.01±0.018 <sup>e</sup>
Mean	12.53±0.195 <sup>a</sup>	12.23±0.210 <sup>b</sup>	12.10±0.210 <sup>c</sup>	

The interaction between WL%, immersion time and SA concentrations treatment mean in a column are significantly different at  $P < 0.05$  (Duncan's multiple rang test). Means followed by the same letter do not differ significantly. Each value represents mean and ± SE.

## DISCUSSION

Postharvest diseases caused by pathogenic fungi are the main key in postharvest losses on guava fruit. Guava fruits contain high levels of nutrients, and their low pH such provided are ideal substrates through postharvest for the development of pathogenic fungi, which in addition to causing fruit deteriorations (Wills & Golding, 2016). The GBS disease caused by *P. capitalensis* is a quiescent infection, that attack immature fruit prior to harvest, under environmental conditions in El-Kharga city, New Valley Governorate-Egypt (Arafat, 2018). The measurement of GBS disease in the three seasons (2015-2017), DI and DS% found different from season to be other. These results harmonized are in agreement with other scientist, who mentioned that, the average incidence of black spot increased with high temperature (Amaral *et al.*, 2006; Escanferla, Moraes, Salaroli, & Massola Jr, 2009). Due to an increasing number of microorganisms are becoming resistant to chemical fungicides treatments. Therefore, search of finding alternative fungicides, safety and environmentally friendly strategy is a major aim of the researchers worldwide. The effect of SA on MLGI% of *P. capitalensis* *in vitro* confirmed at 6 mmol/L completely inhibition. These results agree with those obtained by (Khademi *et al.*, 2012; X. Xu & Tian, 2008; Zhang *et al.*, 2008). Additionally, SA applied can discouragement the growth of other pathogenic molds (Forchetti *et al.*, 2010; Panahirad, Zaare-Nahandi, Safaralizadeh, & Alizadeh-Salteh, 2012). Salicylic acid (SA) is one of the safe, natural compounds applied in the postharvest treatment of fruit. In this study, exogenous application of SA found to decreased DS% at 3 mmol/L to (0%) with three time immersion after three shelf life period at ambient temperature, on guava fruit against *P. capitalensis*. Exogenous application of SA have been reported for control of *Penicillium expansum* in many fruit crops (da Rocha Neto *et al.*, 2016; da Rocha Neto, Maraschin, & Di Piero, 2015; Khademi *et al.*, 2012; X. Xu & Tian, 2008; Zhang *et al.*, 2008) and Alternaria rot (Cao, Yan, Zhao, & Jiang, 2013). Moreover, reports have indicated that SA could build an induced resistance system in fruits and vegetables by inducing the expression of a series of genes, such as those encoding defense related enzymes, including PAL and POD, and pathogenesis related proteins including CHT and GLU (Cao *et al.*, 2013). SA is generally involved in mounting defense mechanisms against biotrophic pathogens, through induced systemic resistance (ISR) and systemic acquired resistance (SAR), respectively (Durrant & Dong, 2004). Data concerning SA immersion treatments of guava fruit showed significant decreased TSS (Brix) and WL% at all concentrations, and time immersion after three shelf life period, compared with control fruit. Apple fruit treated with SA exhibited TSS content than control fruit (M. Kazemi, M. Aran, & Zamani, 2011). In contrast, these results disagree to this, peach fruit treated with SA have highest TSS content compared with control after storage (Tareen, Abbasi, & Hafiz, 2012). SA treatments at 2 mM concentration were highly effective in reducing chilling injury and electrolyte leakage in the husk of pomegranate, as well as ascorbic acid loss (Sayyari, Babalar, Kalantari, Serrano, & Valero, 2009). Meanwhile, addition of SA significantly decreased the level of ROS and lipid peroxidation of cucumber, inhibited its catalase and ascorbate peroxidase activities (Shi & Zhu,

2008). The role of SA decreased WL confirmed in strawberry and peach (Brar, Gupta, & Gill, 2014; Shafiee, Taghavi, & Babalar, 2010; Tareen *et al.*, 2012). Furthermore, SA has shown benefits for human health (Baxter, Graham, Lawrence, Wiles, & Paterson, 2001; Deng, Ruan, Du, Saunders, & Wu, 2001). Overall, data obtained SA treatment against *P. capitalensis* from *in vitro* confirmed complete inhibition. Also, postharvest guava fruit treatments have decreased decay, TSS and WL. Therefore, SA can used in postharvest management of fresh horticultural produce as an alternative to harmful synthetic chemicals to enhance shelf life and ensure food safety.

## CONCLUSION

Postharvest treatment with SA solutions had an overall positive effect on fruit quality of guava during extended shelf life. A delay in ripening, decreased decay incidence, TSS and WL, combined with enhanced quality attributes noticed in treated guava fruit compared with control. The present study demonstrates the efficacy and potential of SA aqueous solutions in preserving the shelf-life of a guava fruit against *Phyllosticta capitalensis* caused GBS disease.

## SIGNIFINANCE STATEMENT

This study discover the applied postharvest treatment with SA that can be beneficial for treatment guava fruits against GBS disease, during shelf life period. This study will help the researchers to uncover the critical areas of natural compounds applied in the postharvest treatment of fruits, that many researchers were not able to explore. Thus, a new theory on SA effective and safe as postharvest treatment of guava fruits against GBS disease may be arrived at.

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## إطالة فترة صلاحية ثمار الجوافة بعد الحصاد بواسطة حمض الساليسيليك ضد مرض البقعة السوداء خالد حسين عرفات قسم امراض النبات- كلية الزراعة- جامعة الوادي الجديد

تعد ثمار الجوافة واحدة من أهم ثمار الفاكهة المنتشرة في المناطق الحارة وشبه الحارة. وهي فاكهة سريعة التلف بعد الحصاد في درجة الحرارة العادية نتيجة نضج الثمار وحوث تغيرات في خواصها الفسيوكيميائية. تهاجم الكائنات الحية الدقيقة ثمار الجوافة، والفطريات أكثرها خطورة، ويعتبر مرض البقعة السوداء في الجوافة من الأمراض الكامنة والتي يسببها الفطر *P. capitalensis* والذي يصيب ثمار الجوافة الغير مكتملة النمو في الحقل، ويؤدي لحدوث خسائر اقتصادية بعد الحصاد. ومنذ ان وجد ان معاملة الثمار بالمبيدات الفطرية بعد الحصاد ضرارة بالإنسان والبيئة، لذلك فإن الهدف الرئيسي للباحثين في جميع انحاء العالم هو البحث عن بدائل للمبيدات الفطرية تكون آمنة للإنسان وصديقة للبيئة. يعتبر حمض الساليسيليك هو احد المركبات الطبيعية والامنة في معاملة الثمار بعد الحصاد. يهدف هذا البحث الى تقييم تأثير التركيزات المختلفة من حمض الساليسيليك على نمو الفطر في المعمل ومعاملة الثمار بعد الحصاد لتحسين العمر الافتراضي لثمار الجوافة. تم تجميع ثمار الجوافة من الأسواق المحلية وتقدير النسبة المئوية لنسبة وشدة الإصابة خلال ثلاثة مواسم متتالية (2015-2017). قدر تأثير التركيزات المختلفة من الحمض على تثبيط النمو الخيطي لميسليوم الفطر في المعمل. عوملت ثمار الجوافة بالغمر في خمس تركيزات مختلفة للحمض، والغمر في ثلاث فترات زمنية مختلفة، ثم الحفظ على درجة حرارة المعمل ثلاث فترات زمنية بعد المعاملة. تم تقدير النسبة المئوية للشدة المرضية، والمواد الصلبة الذائبة الكلية، ونسبة فقد في الوزن لكل فترة حفظ من النتائج المتحصل عليها وجد ان افضل تركيز لتثبيط النمو الفطري في المعمل كان 6 ملليمول/لتر، وكان افضل تركيز لمعاملة الثمار 3ملليمول/لتر لخفض شدة المرض على الثمار، ولقد اعطت فترات غمر الثمار المختلفة تأثيرا معنويا وذلك على فترات الحفظ المختلفة. اوضحت النتائج ان معاملة ثمار الجوافة بحمض الساليسيليك اعطت تأثيرا معنويا في تحسين جودة الثمار اثناء حفظها، وخفض الشدة المرضية، والمواد الصلبة الذائبة ونسبة فقد في وزن الثمار، وذلك مقارنة بالثمار الغير معاملة. اثبتت الدراسة الحالية فعالية حمض الساليسيليك في الحفاظ على العمر الافتراضي لثمار الفاكهة سريعة التلف مثل ثمار الجوافة.